Cont'd

41. (new) A process according to claim 40 wherein said protein precursor is chosen among: preproenzymes, zymogens, matrix metallo proteases, factors belonging to the cascade of the complement system and prohormones.

42. (new) A process according to claim 41 wherein said pre-proenzyme is pre-prourokinase or prourokinase and said mature recombinant protein is tc-uPA.

43.(new) A process for the production of mature recombinant tc-uPA HMW and LMW from a eukaryotic cell line genetically transfected with a cDNA sequence encoding for the human pre-prourokinase, comprising:

a) incubating said cell line in a cell culture medium wherein alkanoic acids, their derivatives or salts therof have been added for a time of at least 24 hours;

b) recovering a cell culture supernatant;

c) performing a ion exchange chromatography on the cell culture supernatant;

d) releasing LMW tc-uPA by addition of a buffer solution with a pH value comprised between 5.5 and 6.5, comprising a monovalent ion in concentration comprised between 200 and 300 mM and optionally further purify LMW tc-uPA by benzamidine chromatography;

e) releasing the HMW tc-UPA by addition of a buffer solution with a pH value comprised between 6-7.5, comprising monovalent ions in concentration of at least 400 mM and optionally further purify HMW tc-uPA by benzamidine chromatography.

44. (new) A process according to claim 43 wherein said alkanoic acids and/or their salts and/or derivatives thereof are chosen among: butyric acid sodium butyrate, sodium propionate, magnesium butyrate, tributyrin and phenyl-butyrate.

45. (new) Process according to claim 44 wherein said alkanoic

acids are in concentration comprised between 0.1 mM and 20 mM.

cont'd

46.(new) A process according to claim 43 wherein said eukaryotic cell line is a mammalian cell line chosen among: HEK-293, CV-1, COS, BSC-1, MDCK, A-431, CHO, BHK, CHO-Messi.

47.(new) A process according to claim 46 wherein said eukaryotic cell line is chosen between CHO and CHO Messi.

48. (new) A process according to claim 43 wherein said temperature of incubation in step a) is comprised between 30°C and 37°C.

49. (new) A process according to claim 48 wherein said temperature is comprised between 33°C and 35°C.

50. (new) A process according to claim 43 wherein said time of incubation in step a) is comprised between 48 and 200 hours.

51. (new) A process according to claim 50 wherein said time is comprised between 72 and 150 hours.

52. (new) A process according to claim 43 wherein said cell culture medium is serum-free.

53. (new) A process according to claim 43 wherein the supernatant recovered in step b) is acidified to pH values comprised between 5 and 5.8, a non-ionic detergent is added and the supernatant is filtered.

54. (new) A process according to claim 43 wherein the benzamidine chromatography in step d) comprises the following steps:

d1) contacting the released LMW tc-uPA containing solution obtained in step d), with a benzamidine column, at pH values comprised between 6 and 8;

d2) releasing the tc-uPA LMW with a buffer solution with

pH values comprised between 3.8 and 4.2 further comprising monovalent ions in concentration comprised between 300 mM and 500mM;

d3) further optionally contacting the released tc-uPA LMW with a gel-filtration column and releasing the LMW tc-uPA with a low-salt solution buffer at a pH comprised between 4 and 7.

55. (new) A process according to claim 43 wherein the benzamidine chromatography in step e) comprises the following steps:

e1) contacting the released HMW tc-uPA containing buffer solution in step e) with a benzamidine column, at pH values comprised between 6.2 and 6.8;

e2) releasing the tc-uPA HMW with a buffer solution having a pH value comprised between 3.8 and 4.2, further comprising monovalent ions in concentration comprised between 300 and 500mM;

e3) further optionally contacting the released tc-uPA HMW with a gel-filtration column and releasing of the HMW tc-uPA with a low-salt solution buffer at pH values comprised between 4 and 7.

56. (new) Cell culture supernatant obtainable by the process of claim 43, step b).

57. (new) Isolated recombinant tc-uPA LMW obtainable by the process of claim 43, step d).

58. (new) Isolated recombinant tc-uPA HMW obtainable by the process of claim 43, step e).

59. (new) Isolated purified recombinant LMW tc-uPA obtainable by the process of claim 54.

60. (new) Isolated purified recombinant HMW tc-uPA obtainable by the process of claim 55.

61. (new) Pharmaceutical compositions comprising as an active agent the recombinant LMW tc-uPA according to claim 59.

62. (new) Pharmaceutical compositions comprising as an active agent the isolated purified recombinant HMW tc-uPA according to claim 60.

63. (new) A method for the treatment of thromboembolytic disorders wherein LMW tc-uPA according to claim 59 is used.

64.(new) A method for the treatment of thromboembolytic disorders wherein HMW tc-uPA according to claim 60 is used.

65. (new) A method according to claim 63 wherein said disorders are chosen among: peripheral arterial occlusion (PAOD), catheter clearance, pulmonary embolism, deep venous thrombosis.

66. (new) A method according to claim 64 wherein said disorders are chosen among: peripheral arterial occlusion (PAOD), catheter clearance, pulmonary embolism, deep venous thrombosis.

67. (new) A method for the treatment of myocardial infarction wherein LMW tc-uPA according to claim 59 is used.

68. (new) A method for the treatment of myocardial infarction wherein HMW tc-uPA according to claim 60 is used.

Kindly cancel claims 2-39.

REMARKS

In response to the restriction request, the applicant elects Group I, claims 1-18, with traverse.

The Applicant respectfully traverses the restriction requirement because a single inventive concept underlies at